AbFlex Cas9 antibody (rAb)

Catalog No.: RA9042

Basic Information

Molecular weight
160 kDa

Category
Recombinant antibody

Applications
ChIP, ChIP-Seq, ELISA, WB

Cross-Reactivity
Human

Background

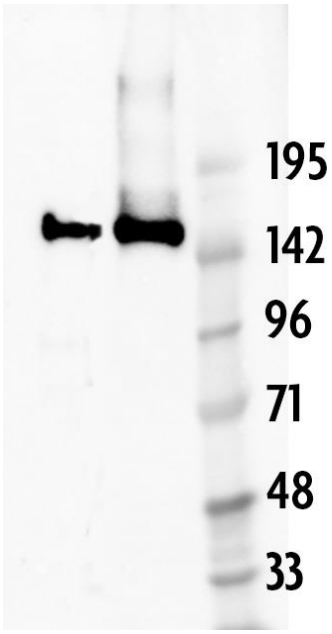
AbFlex antibodies are recombinant antibodies (rAbs) that have been generated using defined DNA sequences to produce highly specific, reproducible antibodies. Each AbFlex antibody contains a 6xHis Tag, a Biotinylation Tag for enzymatic biotin conjugation using the biotin ligase, BirA, and a sortase recognition motif (LPXTG) to attach a variety of labels directly to the antibody including fluorophores, enzymatic substrates (HRP, AP), peptides, drugs as well as solid supports. AbFlex Cas9 antibody was expressed as full-length IgG with mouse immunoglobulin heavy and light chains (IgG2a isotype) in mammalian 293 cells. Cas9 is a nuclease from Streptococcus pyogenes that can be targeted to particular DNA sequences through a guide RNA that results in double-stranded breaks in DNA. Cas9 is part of the CRISPR/Cas9 gene-editing system that can create a DNA break at a specific location with the genome. CRISPR (clustered regularly interspaced short palindromic repeat) is an adaptive immune system that provides protection against mobile genetic elements (viruses, transposable elements and conjugative plasmids). CRISPR clusters contain spacers, sequences complementary to antecedent mobile elements, and target invading nucleic acids.

Recommended Dilutions

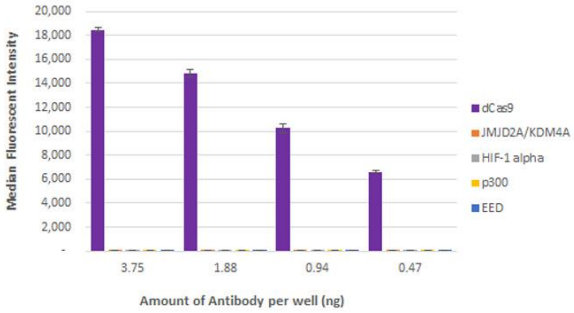
ChIP-Seq	4 μg
ELISA	9 - 75 ng/ml
WB	2 μg/ml

Product Information

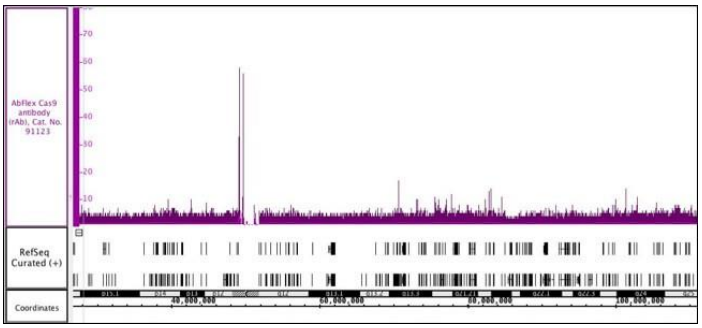
Source	Mouse
Isotype	IgG2a
Purification	Ni-NTA
Storage buffer	Purified IgG in 200 mM Hepes pH 7.5, 100 mm NaCl, 50 mM NaOAc, 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.
Storage Conditions	Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage.



AbFlex Cas9 antibody (rAb) tested by Western blot. 50 ng recombinant Cas9 protein was probed with 0.5 ug/ml AbFlex Cas9 antibody (Lane 1). HEK293T cells were transfected with a plasmid containing a mammalian expression construct for dCas9 (S. pyogenes). Chromatin was prepared 48 hours post-transfection, boiled and 15 ul (~200,000 cell equivalents) was run on a SDS-PAGE gel and probed with 2 ug/ml AbFlex Cas9 antibody (Lane 2). A molecular weight marker was run in Lane 3.



AbFlex Cas9 antibody (rAb) tested by bead-based specificity analysis. Luminex bead-based specificity analysis was used to confirm the specificity of AbFlex Cas9 antibody (rAb) antibody for Cas9. Various proteins were conjugated to MagPlex Luminex beads and incubated with various amounts of AbFlex Cas9 antibody (rAb). Protein-bound antibody was detected with anti-mouse IgG-Phycoerythrin and read in a Luminex instrument. Luminex is a registered trademark of Luminex Corporation.



AbFlex Cas9 recombinant antibody (rAb) tested by ChIP-Seq Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT High Sensitivity Kit (Cat. No. 53040) with 30 ug of Jurkat cell chromatin and 4 ug of AbFlex Cas9 antibody. ChIP DNA was sequenced on the Illumina NextSeq and 8 million sequence tags were mapped to identify binding sites on chromosome 4.